

ER TA Reporter Gene Assays Using Yeast Cells

	Arnold et al. (1996)	Beresford et al. (2000)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i> *	<i>S. cerevisiae</i> *
Strain	BJ2407	n.p.
Stable Transfection of Cells with Plasmids		
ER expression vector	PSCW231-hER	hER -ppk
ER source	human	human
Reporter vector	YRPE2 LacZ	vit2ERE-LacZ
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	DMSO	ethanol
Range of test substance concentrations	0.001 to 10,000 nM	n.p.
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	At least 2
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	1000 nM	4.88 pmoles - 2 nmoles
Incubation time of test substance	overnight	3 days
Measured as (e.g., color change, growth)	ONPG color change	CPRG color change
Antagonism		
Reference ligand		17 -Estradiol
Final concentration of reference ligand		2.5 x 10 ⁻¹⁰ M
Measured as (e.g., color change)		CPRG color change

Abbreviations: DMSO = dimethyl sulfoxide;
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* Species name not provided in publication,
but likely *S. cerevisiae*

ER TA Reporter Gene Assays Using Yeast Cells

	Chen et al. (1997)	Chen et al. (1997)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i> *	<i>S. cerevisiae</i> *
Strain	939	CTY10-5d
Stable Transfection of Cells with Plasmids		
ER expression vector	pUC19hER	LexA-hER
ER source	human	human
Reporter vector	YRpE2; cyc promotor; ERE-cyc-LacZ	ERE-LacZ
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	DMSO	DMSO
Range of test substance concentrations	n.p.	n.p.
No. of replicates	3-6	3-6
No. of times assay repeated	n.p.	n.p.
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	100 nM	100 nM
Incubation time of test substance	12 hours	12 hours
Measured as (e.g., color change, growth)	ONPG color change	ONPG color change
Antagonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	0.5 nM	1 nM
Measured as (e.g., color change)	ONPG color change	ONPG color change

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	Coldham et al. (1997)	Connor et al. (1996)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i> *	<i>S. cerevisiae</i>
Strain	n.p.	PL3
Stable Transfection of Cells with Plasmids		
ER expression vector	ER-CUP1 MET	YEp10-HEGO
ER source	human	human
Reporter vector	2FR.vit-iso1-cytC-lacZ	URA 3
Endpoint measured	-galactosidase	growth
Other plasmids	none	none
Transfection reagent	n.p.	Lithium acetate
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	n.p.	DMSO
Range of test substance concentrations	n.p.	n.p.
No. of replicates	n.p.	4
No. of times assay repeated	n.p.	3
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	n.p.	1 nM
Incubation time of test substance	18 hours	n.p.
Measured as (e.g., color change, growth)	ONPG color change	growth/no growth
Antagonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	0.001 - 10 µM	1 nM
Measured as (e.g., color change)	-gal induction	cell division

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	De Boever et al. (2001)	Elsby et al. (2001)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	n.p.	n.p.
Stable Transfection of Cells with Plasmids		
ER expression vector	ER (otherwise undefined)	ER (otherwise undefined)
ER source	human	human
Reporter vector	n.p.	Yeast 3 phosphoglycerol promoter with EREs
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	Overnight	n.p.
Transcriptional Activation Assay		
Metabolic activation	yes	yes (2 chems) and no
Metabolic activation source	intestinal bacterial -glycosidase	female Wistar rat liver; female human liver microsomes
Test substance solvent	ethanol	n.p.
Range of test substance concentrations	10 ⁻⁴ to 10 ² mol/L	1 pM to 100 nM
No. of replicates	24 hours: 4 / 3-day: 4-6	2
No. of times assay repeated	n.p.	4-8
Agonism		
Reference ligand	17-Estradiol	17-Estradiol
Final concentration of reference ligand	6.96 nmoles	n.p.
Incubation time of test substance	24 hours / 3 days	3-4 days
Measured as (e.g., color change, growth)	CPRG color change	n.p.
Antagonism		
Reference ligand		17-Estradiol
Final concentration of reference ligand		1x10 ⁻¹⁰ M
Measured as (e.g., color change)		CPRG color change

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	Gaido et al. (1997)	Grauman et al. (1999)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i> *
Strain	BJ3505	188R1
Stable Transfection of Cells with Plasmids		
ER expression vector	CUP1 hER	YEpE12 (HER) + CUP1
ER source	human	human
Reporter vector	ERE-LacZ	YRpE2 (vitellogenin A2ERE; cyc1; lacZ)
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	CUP1 metallothionein promoter	CUP1 promotor
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	methanol	DMSO
Range of test substance concentrations	n.p.	10 pM to 100 nM
No. of replicates	n.p.	n.p.
No. of times assay repeated	3-5	n.p.
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	n.p.	varied
Incubation time of test substance	overnight	4 hours
Measured as (e.g., color change, growth)	ONPG color change	-galactosidase activity
Antagonism		
Reference ligand		17 -Estradiol
Final concentration of reference ligand		n.p.
Measured as (e.g., color change)		-galactosidase activity

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ER TA Reporter Gene Assays Using Yeast Cells

	Harris et al. (1997)	Klotz et al. (1996)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i> *
Strain	n.p.	BJ2407
Stable Transfection of Cells with Plasmids		
ER expression vector	n.p.	PSCW231-hER
ER source	human	human
Reporter vector	vit2ERE-LacZ	YRP2ERE
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	Overnight	Overnight
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	ethanol	DMSO
Range of test substance concentrations	n.p.	n.p.
No. of replicates	n.p.	3
No. of times assay repeated	n.p.	2
Agonism		
Reference ligand	17-Estradiol	17-Estradiol
Final concentration of reference ligand	1 x 10 ⁻⁸ M	0.01 µM
Incubation time of test substance	4-6 days	overnight
Measured as (e.g., color change, growth)	CPRG color change	ONPG color change
Antagonism		
Reference ligand		
Final concentration of reference ligand		
Measured as (e.g., color change)		

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ER TA Reporter Gene Assays Using Yeast Cells

	Lascombe et al. (2000)	Le Guevel and Pakdel (2001)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i> *	<i>S. cerevisiae</i>
Strain	YRG-2	BJ-ECZ
Stable Transfection of Cells with Plasmids		
ER expression vector	p2HG-hER	hER (otherwise undefined)
ER source	human	human
Reporter vector	pLGERE-CYC-1	ERE CYC1
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	Overnight	36 hours
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	Ethanol	DMSO
Range of test substance concentrations	10 pM to 1 µM	10 ⁻¹⁰ to 10 ⁻⁵ mol/L
No. of replicates	3	4
No. of times assay repeated	At least 2	6-9
Agonism		
Reference ligand	17 -Estradiol	Ethinyl estradiol
Final concentration of reference ligand	n.p.	n.p.
Incubation time of test substance	overnight	4 hours
Measured as (e.g., color change, growth)	-gal induction	ONPG color change
Antagonism		
Reference ligand	17 -Estradiol	
Final concentration of reference ligand	n.p.	
Measured as (e.g., color change)	-gal induction	

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	Le Guevel and Pakdel (2001)	Miller et al. (2001)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i> *
Strain	BJ-ECZ	n.p.
Stable Transfection of Cells with Plasmids		
ER expression vector	rtER (otherwise undefined)	ER (otherwise undefined)
ER source	rainbow trout	human
Reporter vector	ERE2-CYC1-LacZ	"expression plasmids" with EREs ppk
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	36 hours	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	DMSO	ethanol
Range of test substance concentrations	10 ⁻¹⁰ to 10 ⁻⁵ mol/L	1 pM to 1 mM
No. of replicates	4	n.p.
No. of times assay repeated	6-12	At least 2
Agonism		
Reference ligand	Ethinyl estradiol	17-Estradiol
Final concentration of reference ligand	n.p.	4.88 pmoles - 2 nmoles
Incubation time of test substance	4 hours	3 days
Measured as (e.g., color change, growth)	ONPG color change	CPRG color change
Antagonism		
Reference ligand		
Final concentration of reference ligand		
Measured as (e.g., color change)		

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ER TA Reporter Gene Assays Using Yeast Cells

	Moffat et al. (2001)	Morito et al. (2001a)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	n.p.	Y190
Stable Transfection of Cells with Plasmids		
ER expression vector	hER (otherwise undefined)	pGBT9-hER
ER source	human	human
Reporter vector	n.p.	n.p.
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	Ethanol	n.p.
Range of test substance concentrations	10 ⁻¹² to 0.1 M	0.01 pM to 0.1 mM
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	n.p.
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	n.p.	n.p.
Incubation time of test substance	3 days	n.p.
Measured as (e.g., color change, growth)	-gal induction	-gal induction
Antagonism		
Reference ligand	17 -Estradiol	
Final concentration of reference ligand	0.005 nM	
Measured as (e.g., color change)	-gal induction	

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ER TA Reporter Gene Assays Using Yeast Cells

	Morito et al. (2001a)	Morito et al. (2001b)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	Y190	Y190
Stable Transfection of Cells with Plasmids		
ER expression vector	pGBT9-hER	pGBT9-hER
ER source	human	human
Reporter vector	n.p.	n.p.
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	n.p.	n.p.
Range of test substance concentrations	0.01 pM to 0.1 mM	1 pM to 0.1 mM
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	n.p.
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	n.p.	n.p.
Incubation time of test substance	n.p.	n.p.
Measured as (e.g., color change, growth)	-gal induction	-gal induction
Antagonism		
Reference ligand		17 -Estradiol
Final concentration of reference ligand		1 nM
Measured as (e.g., color change)		-gal induction

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ER TA Reporter Gene Assays Using Yeast Cells

	Morito et al. (2001b)	Odum et al. (1999)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	Y190	n.p.
Stable Transfection of Cells with Plasmids		
ER expression vector	pGBT9-hER	hER
ER source	human	human
Reporter vector	n.p.	n.p.
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	n.p.	ethanol
Range of test substance concentrations	1 pM to 0.1 mM	1 pM to 1mM
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	n.p.
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	n.p.	n.p.
Incubation time of test substance	n.p.	4 days
Measured as (e.g., color change, growth)	-gal induction	color change (otherwise undefined)
Antagonism		
Reference ligand	17 -Estradiol	
Final concentration of reference ligand	1 nM	
Measured as (e.g., color change)	-gal induction	

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	Petit et al. (1997)	Petit et al. (1999)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	BJ-ECZ	BJ-ECZ
Stable Transfection of Cells with Plasmids		
ER expression vector	YEprtER	YEprtER
ER source	rainbow trout	rainbow trout
Reporter vector	2ERE-CYC1-LacZ	2ERE-CYC1-LacZ
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	n.p.	n.p.
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	ethanol or DMSO	ethanol or DMSO
Range of test substance concentrations	10 pM to 0.1 mM	10 pM to 0.1 mM
No. of replicates	n.p.	n.p.
No. of times assay repeated	At least 3	At least 2
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	10 nM	10 nM
Incubation time of test substance	4 hours	4 hours
Measured as (e.g., color change, growth)	-gal induction	-gal induction
Antagonism		
Reference ligand		
Final concentration of reference ligand		
Measured as (e.g., color change)		

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but likely *S. cerevisiae*

ER TA Reporter Gene Assays Using Yeast Cells

	Rajapakse et al. (2001)	Ramamoorthy et al. (1997a)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i> *	<i>S. cerevisiae</i> *
Strain	n.p.	BJ2168
Stable Transfection of Cells with Plasmids		
ER expression vector	hER	CUP1
ER source	human	mouse
Reporter vector	n.p.	vit ERE1-lacZ
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	Lithium acetate
Plating time prior to treatment with test substance	Overnight	Overnight
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	Ethanol	DMSO
Range of test substance concentrations	10 ⁻⁵ to 10 ² µM	250 nM to 25 µM
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	4
Agonism		
Reference ligand	17 -Estradiol	Diethylstilbestrol
Final concentration of reference ligand	n.p.	1 and 10 nM
Incubation time of test substance	72 hours	2.5 hours or 16 hours
Measured as (e.g., color change, growth)	ONPG color change	-gal induction
Antagonism		
Reference ligand		Diethylstilbestrol
Final concentration of reference ligand		10 nM
Measured as (e.g., color change)		-gal induction

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	Ramamoorthy et al. (1997a)	Ramamoorthy et al. (1997b)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i> *	<i>S. cerevisiae</i>
Strain	BJ2407	BJ3505
Stable Transfection of Cells with Plasmids		
ER expression vector	YePE10	CUP1-hER
ER source	human	human
Reporter vector	YRPE2 vit 2 CYC1	n.p.
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	Lithium acetate	n.p.
Plating time prior to treatment with test substance	Overnight	Overnight
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	DMSO	DMSO
Range of test substance concentrations	n.p.	0.1 pM to 10 µM
No. of replicates	n.p.	3
No. of times assay repeated	n.p.	n.p.
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	10 nM	.001 µM
Incubation time of test substance	24 hours	overnight
Measured as (e.g., color change, growth)	-gal induction	ONPG color change
Antagonism		
Reference ligand	17 -Estradiol	
Final concentration of reference ligand	10 nM	
Measured as (e.g., color change)	-gal induction	

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ER TA Reporter Gene Assays Using Yeast Cells

	Routledge & Sumpter (1996)	Routledge and Sumpter (1997)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	n.p.	n.p.
Stable Transfection of Cells with Plasmids		
ER expression vector	hER	hER
ER source	human	human
Reporter vector	vit 2 -gal	n.p.
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	24 hours	24 hours
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	ethanol	ethanol
Range of test substance concentrations	60 nM to 5 mM	1 pM to 10 mM
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	1
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	3000 ng/L	10 nM
Incubation time of test substance	3 days	3 days
Measured as (e.g., color change, growth)	CPRG color change	CPRG color change
Antagonism		
Reference ligand		none
Final concentration of reference ligand		n.p.
Measured as (e.g., color change)		CPRG color change

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ER TA Reporter Gene Assays Using Yeast Cells

	Tran et al. (1996)	Tran et al. (1996)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	ER (wt)	ER179C
Stable Transfection of Cells with Plasmids		
ER expression vector	hER	hER
ER source	human	human
Reporter vector	n.p.	n.p.
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	Overnight	Overnight
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	DMSO	DMSO
Range of test substance concentrations	n.p.	n.p.
No. of replicates	n.p.	n.p.
No. of times assay repeated	n.p.	n.p.
Agonism		not done
Reference ligand	17 -Estradiol	
Final concentration of reference ligand	0.5 nM	
Incubation time of test substance	12 hours	
Measured as (e.g., color change, growth)	growth; ONPG color change	
Antagonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	0.5 nM	0.5 nM
Measured as (e.g., color change)	ONPG color change	growth; ONPG color change

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n.a. = not applicable; n.p. = not provided.

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ER TA Reporter Gene Assays Using Yeast Cells

	Vinggaard et al. (1999)	Vinggaard et al. (2000)
Characteristics of Yeast		
Species	<i>S. cerevisiae</i>	<i>S. cerevisiae</i>
Strain	n.p.	n.p.
Stable Transfection of Cells with Plasmids		
ER expression vector	hER	hER
ER source	human	human
Reporter vector	vit 2 -gal	vit 2 -gal
Endpoint measured	-galactosidase	-galactosidase
Other plasmids	none	none
Transfection reagent	n.p.	n.p.
Plating time prior to treatment with test substance	24 hours	24 hours
Transcriptional Activation Assay		
Metabolic activation	no	no
Metabolic activation source	n.a.	n.a.
Test substance solvent	ethanol	ethanol
Range of test substance concentrations	0.2 µM to 550 µM	0.2 µM to 550 µM
No. of replicates	n.p.	n.p.
No. of times assay repeated	3	2
Agonism		
Reference ligand	17 -Estradiol	17 -Estradiol
Final concentration of reference ligand	0.2 - 500 pM	0.24 - 500 pM
Incubation time of test substance	4 days	4 days
Measured as (e.g., color change, growth)	CPRG color change	CPRG color change
Antagonism		
Reference ligand		
Final concentration of reference ligand		
Measured as (e.g., color change)		

Abbreviations: DMSO = dimethyl sulfoxide;
n.a. = not applicable; n.p. = not provided.

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ER TA Reporter Gene Assays Using Yeast Cells

	Yoshihara et al. (2001)
Characteristics of Yeast	
Species	<i>S. cerevisiae</i> *
Strain	n.p.
Stable Transfection of Cells with Plasmids	
ER expression vector	hER
ER source	human
Reporter vector	lacZ
Endpoint measured	-galactosidase
Other plasmids	none
Transfection reagent	n.p.
Plating time prior to treatment with test substance	n.p.
Transcriptional Activation Assay	
Metabolic activation	yes
Metabolic activation source	male Wistar rat
Test substance solvent	ethanol
Range of test substance concentrations	n.p.
No. of replicates	At least 2
No. of times assay repeated	n.p.
Agonism	
Reference ligand	17 -Estradiol
Final concentration of reference ligand	0.05 µM
Incubation time of test substance	1 hours (+S9); 24-48 hours (-S9)
Measured as (e.g., color change, growth)	-gal induction
Antagonism	
Reference ligand	not done
Final concentration of reference ligand	
Measured as (e.g., color change)	

Abbreviations: DMSO = dimethyl sulfoxide;
n.a. = not applicable; n.p. = not provided.

* Species name not provided in publication,
but likely *S. cerevisiae*